



Citation: Zhou H, Zhao WZ, He ZB, *et al.*, 2018. Stable isotopes reveal varying water sources of *Caragana microphylla* in a desert-oasis ecotone near the Badain Jaran Desert. *Sciences in Cold and Arid Regions*, 10(6): 0458–0467. DOI: 10.3724/SP.J.1226.2018.00458.

Stable isotopes reveal varying water sources of *Caragana microphylla* in a desert-oasis ecotone near the Badain Jaran Desert

Hai Zhou¹, WenZhi Zhao^{1*}, ZhiBin He¹, Heng Ren^{1,2}

1. Linze Inland River Basin Research Station, Key Laboratory of Ecohydrology of Inland River Basin, Northwest Institute of Eco-Environment and Resources, Chinese Academy of Sciences, Lanzhou, Gansu 730000, China

2. Lanzhou Information Center, Chinese Academy of sciences, Lanzhou, Gansu 730000, China

*Correspondence to: WenZhi Zhao, Linze Inland River Basin Research Station, Key Laboratory of Ecohydrology of Inland River Basin, Northwest Institute of Eco-Environment and Resources, Chinese Academy of Sciences. No. 320, West Donggang Road, Lanzhou, Gansu 730000, China. Tel: +86-931-4967137; E-mail: zhaowzh@lzb.ac.cn

Received: June 6, 2018 Accepted: July 13, 2018

ABSTRACT

Understanding the variation in a plant's water sources is critical to understanding hydrological processes in water-limited environments. Here, we measured the stable-isotope ratios ($\delta^{18}\text{O}$) of xylem water of *Caragana microphylla*, precipitation, soil water from different depths, and groundwater to quantitatively analyze the proportion of water sources for the shrub. We found that the water sources of *C. microphylla* differed with the plant's ages and the seasons. The main water source for young shrubs was upper-soil water, and it showed significant changes with seasonal precipitation inputs. In summer, the proportion contributed by shallow water was significantly increased with increased precipitation inputs. Then, the contribution from shallow-soil water decreased with the decline in precipitation input in spring and autumn. However, the adult shrubs resorted to deep-soil layers and groundwater as the main water sources during the whole growing season and showed much less seasonal variation. We conclude that the main water source of the young shrubs was upper-soil water and was controlled by precipitation inputs. However, once the shrub gradually grew up and the roots reached sufficient depth, the main water sources change from the upper-soil layer recharged by precipitation to deep-soil water and groundwater, which were relatively stable and abundant in the desert ecosystem. These results also suggest that desert shrubs may be able to switch their main water sources to deep and reliable water sources as their age increases, and this adjustment to water availability carries significant importance for their acclimation to the desert habitat.

Keywords: water-use pattern; oxygen stable isotope; water sources; *Caragana microphylla*

1 Introduction

In arid and semi-arid environments, water is the principal limiting resource that determines a plant's survival and growth, its carbon fixation, and vegetation distribution (Fay *et al.*, 2003; Bunker *et al.*, 2005). Precipitation and groundwater are the main water sources for plants (Ehleringer *et al.*, 1991;

Dawson *et al.*, 2002). Plant activity is tightly coupled to water availability, and water-use strategies influence plant species' responses to fluctuating environmental conditions (Xu and Li, 2006). In arid and semiarid regions, the uneven distribution of precipitation brings about extreme changes in soil water over both space and time (Schwinning and Ehleringer, 2001; Loik *et al.*, 2004; Trogisch *et al.*, 2016). In re-

gions where evaporation demand exceeds precipitation and the upper-soil water tends to be unavailable, plants are likely to regulate their water-use patterns; and their roots have to acquire any remaining soil moisture (Lilley and Fukai, 1994; West *et al.*, 2008; Grossiord *et al.*, 2017). Therefore, patterns in the water use of desert plants play a crucial role in shaping plant adaptation in water-use strategies and in determining the composition of plant communities.

Plants in desert environments often rely on access to deep and moist soil layers to withstand seasonal heat waves and droughts (Eggemeyer *et al.*, 2009; Rossatto *et al.*, 2012; Grossiord *et al.*, 2017). Water-use patterns can also vary following changes in precipitation source (Snyder and Williams, 2003; Barbeta *et al.*, 2015). This ability to shift among main water sources based on available soil moisture depends on the depth and distribution of functional roots (Dawson and Pate, 1996). Desert plants exploit deep, dependable water sources, such as deep-soil water or groundwater, making it possible for some plants to survive long periods without rain or to overcome extreme drought conditions (Wu *et al.*, 2014; Dai *et al.*, 2015). Some plants also have a dimorphic rooting system, with roots distributed in shallow and deep soils, which can enable the uptake of seasonal rain and simultaneously allow the extraction of water from deeper soils that have been charged during previous seasons (Ehleringer and Dawson, 1992; Dawson and Pate, 1996). Therefore, species adapted to recurrent droughts by maintaining these characteristic rooting systems may have a competitive advantage in the arid ecosystem (Zhou *et al.*, 2015; Grossiord *et al.*, 2017). However, changes in water-use patterns by plants might be expected due to the different above- and belowground microenvironmental characteristics experienced by plants of different ages and sizes in arid and semiarid regions (Donovan and Ehleringer, 1991; Matzner *et al.*, 2003; Song *et al.*, 2016). Adult plants may have deeper roots and, therefore, greater access to deeper water sources than young plants (Drake *et al.*, 2011; Kerhoulas *et al.*, 2013), thereby avoiding or minimizing the effects of drought in arid and semiarid regions.

An analysis of the stable-isotope composition of water provides a powerful tool for expounding the movement of water through ecosystems (Kendall and McDonnell, 1998), and a plant's main water sources can be determined by comparing the stable-isotope ratios of all potential water sources (such as precipitation, soil water from varying depths, groundwater) with those of water extracted from the plant xylem (Ehleringer and Dawson, 1992; Dawson *et al.*, 2002; Eggemeyer *et al.*, 2009; Zhou *et al.*, 2017). The stable isotope can be applied to water-acquisition studies because no fractionation occurs during water absorption

by the roots of terrestrial plants (Mensforth *et al.*, 1994; Dawson *et al.*, 2002). If the hydrology and oxygen isotopic composition of potential water sources are analyzed before the water has been exposed to evaporative processes, this isotopic composition is an integrated measure of overall water uptake, reflecting the various zones and depths from which the plant is currently extracting water (Ehleringer and Dawson, 1992; Dawson *et al.*, 2002). As such, stable-isotope analyses of a plant's water sources provide a powerful tool to improve our understanding of the plant's active rooting zone and water-uptake processes, and also provide insight into the role of water in influencing the ecological and physiological processes (Ehleringer and Dawson, 1992). This technique has been widely applied in the field of ecohydrology, especially in arid and semiarid regions (Dawson *et al.*, 2002; Eggemeyer *et al.*, 2009; Wu *et al.*, 2014; Garcia-Fornier *et al.*, 2016).

C. microphylla, as a typical xerophyte, has physiological and morphological traits that allow it to survive in the frequent aridity, torridity, and other environmental stresses in arid regions (Su and Zhao, 2003; Guan *et al.*, 2015). But the influence of variability in water sources on *C. microphylla* growth is not well understood. Lack of quantitative research on plants' water sources is mainly due to the unpredictable nature of precipitation and the complexity of its function. Based on the stable-isotope technology, the objectives of the current study were to understand how the shrub uses precipitation inputs, which vary strongly with the season, and the groundwater, and how this use changes with as the shrub ages.

2 Materials and methods

2.1 Study site

This study was conducted at the Linze Inland River Basin Research Station, Chinese Academy of Sciences (39°21'N, 100°07'E, 1,374 m a.s.l.), which is located at the Linze oasis fringe and near the Badain Jaran Desert (Figure 1). This region experiences an extremely arid climate. The mean annual precipitation is 116.8 mm (mean for the period 1965–2000), and more than 80% of the precipitation occurs between May and September. But the mean annual open-water evaporation is 2,390 mm, exceeding the mean annual precipitation. The prevailing wind direction is northwest, with a 3.2 m/s mean annual wind velocity. Soils are characterized by a coarse texture and loose structure, and are very susceptible to wind erosion. The groundwater table depth is about 5.0 m, and its seasonal change scale is small. The landscape includes fixed, semifixed, semimobile, and mobile dunes. Native plants on the fixed and semifixed dunes

include shrubs (e.g., *C. microphylla*, *Haloxylon ammodendron*, *Tamarix ramosissima*, and *Nitraria sphaerocarpa*) and herbaceous species (e.g., *Bassia dasyphylla*, *Halogeton arachnoideus*, *Suaeda glauca*, and *Agriophyllum squarrosum*).

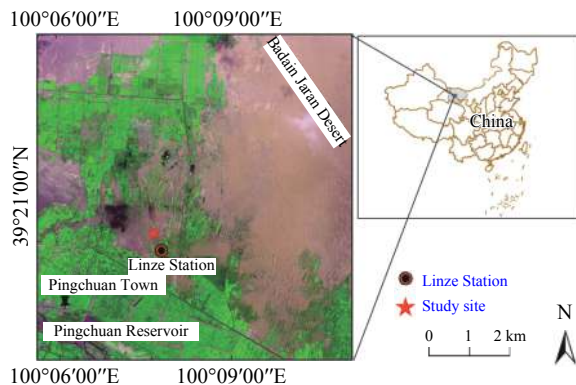


Figure 1 Location of the study site

2.2 Sample collection

Precipitation samples were collected by a simple rain isotopic-sample collector during almost all precipitation events. On each sampling date, we collected groundwater samples as well. Both precipitation and groundwater samples were filtered using a 0.22- μm filter and immediately enclosed in glass vials, wrapped in Parafilm, then refrigerated at 2 °C. Precipitation amounts and temperatures in the study area were recorded by a weather station at the Linze Inland River Basin Research Station. Plant and soil sampling was conducted simultaneously to characterize seasonal water-isotope signatures from April to October 2014. At each site, three healthy shrubs with mean crown breadth of 250 cm were selected; and five small twigs (diameter 0.2–0.5 cm, length 4–5 cm) were collected at the base of the live crown of each study plant at monthly intervals. Phloem from the twigs was immediately removed and placed in glass vials and sealed with a screw-lid and Parafilm wrap, then placed in a frozen box for transportation to the laboratory. For the soil samples, three replicates at soil depths of 0 to 300 cm were obtained by hand auger under the canopy of the study plants on each sampling date; from depths as 0 to 100 cm, collected at 10-cm intervals, and from depths as over 100 to 300 cm at 20-cm intervals. Soil samples were separated into two parts, for stable-isotope analyses and water-content determination. One subsample was immediately double-bagged in glass vials, refrigerated, and transported to the laboratory for stable-isotope analyses; the other subsample was sealed in tin cups

for measuring soil-water content (SWC) by the oven-drying method.

2.3 Isotopic analyses

Using cryogenic vacuum distillation, the water of the plant xylem and soil was extracted (Ehleringer *et al.*, 2000) and stored in sealed glass vials at 2 °C before being measured. All water samples were measured by infrared spectroscopy (IRIS) analyzer—the Liquid Water Isotope Analyzer (LWIA, 912-0008-1001, Los Gatos Research Inc., Mountain View, CA, USA). IRIS-extracted stem water of some species may contain compounds that interfere with accurate isotope ratio measurements; these potential contaminants were identified and quantified with the spectral contamination identifier (LWIA) post-processing software, and the isotope values of contaminated water samples were corrected (Schultz *et al.*, 2011). For details of the procedure, refer to Wu *et al.* (2014). Because hydrogen isotopic fractionation has been observed, but oxygen isotopic fractionation is negligible during water uptake by certain halophytic or xerophytic plants (Ellsworth and Williams, 2007), we used only oxygen isotopes to determine plant-water sources. All stable isotope values are reported in "delta" notation, which expresses the isotopic composition of a material relative to that of an accepted standard (Vienna Standard Mean Ocean Water, V-SMOW) on a per mil (‰) basis:

$$\delta^{18}\text{O} = \left(\frac{R_{\text{sample}}}{R_{\text{standard}}} - 1 \right) \times 1000\text{‰} \quad (1)$$

where R_{sample} and R_{standard} are the oxygen stable-isotopic compositions ($^{18}\text{O}/^{16}\text{O}$ molar ratio) of the sample and standard water (Standard Mean Ocean Water, SMOW).

2.4 Data analyses

Isotopic composition of xylem water was compared with that of potential water sources, with five potential water sources being tested in our research: precipitation, shallow-soil water (0–50 cm), mid-depth-soil water (50–150 cm), deep-soil water (150–300 cm), and groundwater. The contribution of each source to the plant was calculated by the IsoSource model (Phillips and Gregg, 2003). Independent-sample *t*-tests were used to test the differences in $\delta^{18}\text{O}$ values of xylem water between the young and adult *C. microphylla*. Significance was determined at the 95% confidence level ($\alpha = 0.05$). All statistical analyses were performed with SPSS software (version 17.0, SPSS Inc., Chicago, IL, USA).

3 Result

3.1 Stable-isotope variations in precipitation and soil water

In the study area, the total precipitation in 2014 was 102.8 mm, 11.7% lower than the long-term mean annual precipitation. During the measurement period (from April to October 2014), total precipitation was 99.6 mm, accounting for 96.8% of that year's precipitation. There was no precipitation between January and March. Precipitation events less than or equal to 5 mm accounted for 57.8% of the total precipitation and 87.8% of the events, and precipitation events of 5–10 mm accounted for 34.9% of the total precipitation and 14.6% of the events. On July 22, the precipitation event (18 mm) in 2014 happened and was the only event greater than 10 mm (Figure 2). The stable-isotope ratios ($\delta^{18}\text{O}$) of precipitation changed significantly during the precipitation events, ranging from -7.03‰ to 8.85‰ (Figure 2). The stable isotope of summer precipitation in $\delta^{18}\text{O}$ values was negatively correlated with precipitation amount ($r = -0.732$, $p = 0.039$, $n = 15$), exhibiting a significant effect of precipitation amount (Dansgaard, 1964). The stable-isotope ratios of precipitation in spring (April to May) and autumn (September to October) were lower than those in summer (June to August).

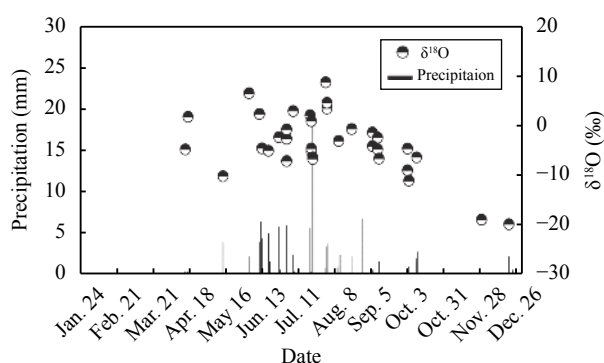


Figure 2 $\delta^{18}\text{O}$ values for rainwater and the corresponding precipitation events, with the average daily temperature between April and October 2014

3.2 Stable-isotope variations in soil water

During the measurement period in 2014, soil waters (at depths of 0–100 cm) had highly variable stable-isotope ratios ($\delta^{18}\text{O}$) due to inputs from rain with variable isotope signatures and from the evaporative enrichment effect. Especially for the upper 40 cm of the soil layer, the stable-isotope ratio ($\delta^{18}\text{O}$) for soil water was more affected by enrichment in summer and autumn than in spring, and was depleted as the depth increased (Figure 3a). However, the

stable-isotope ratio ($\delta^{18}\text{O}$) of the soil water at depths of 40–60 cm and 60–100 cm was significantly more affected by enrichment in spring and summer than in autumn. Perhaps the summer precipitation infiltrated into the deeper soil profile and changed the stable isotope. For the SWC, the upper-soil layer (0–40 cm) changed more extensively than that of the deeper-soil layers, due to the effect of precipitation and soil evapotranspiration. The SWC in the 0–20 cm soil profile was significantly lower than that at 20–40 cm and 40–60 cm in May and June, but it gradually increased from July to September due to the increase in precipitation. At the depth of 50–100 cm, SWC exhibited mild seasonal fluctuations and significantly increased with depth at the site (Figure 3).

3.3 Stable-isotope variations in shrub xylem water

The stable-isotope ratios ($\delta^{18}\text{O}$) of *C. microphylla* xylem water exhibited high variability within seasons and across ages (Figure 4). During the measurement period (April to October 2014), young shrubs had stable-isotope ratios ($\delta^{18}\text{O}$) of xylem water ranging from -1.39‰ to 5.66‰ . Seasonal change in the stable-isotopic ratios was nearly synchronous with that in precipitation (Figure 4), showing precipitation influenced water sources of the young shrubs. For the adult shrubs, the stable-isotope ratios ($\delta^{18}\text{O}$) were lowest, and fluctuated mildly with seasons, and were similar to the ratios of deep-soil water and groundwater. The stable-isotope ratios ($\delta^{18}\text{O}$) between the young and adult shrubs showed significant differences ($P < 0.01$), probably indicating that the shrub's water sources changed from shallow-soil water recharged by precipitation, to deep water (including deep-soil water and groundwater) as the plant grew older.

The stable-isotope ratios ($\delta^{18}\text{O}$) of soil water at the study site changed with the seasons and exhibited a monotonic decline along the depth of the soil profile (Figure 5). The upper-soil layers were consistently more enriched with the heavier isotope than the lower-soil horizons (Figures 3a, 5). The stable-isotope ratios ($\delta^{18}\text{O}$) of soil water at 40–150 cm showed relatively mild fluctuations with the seasons ($P < 0.05$), while those below 150 cm had no significant seasonal variation ($P > 0.05$). Deep soil was usually more depleted in isotope ratios than upper soils but was more enriched than groundwater. Compared with the soil water, the groundwater exhibited relatively steady isotope values (-8.17‰ to -8.91‰ for $\delta^{18}\text{O}$) during the measurement period. The stable-isotope ratios ($\delta^{18}\text{O}$) of the xylem water of the young shrubs matched that of the soil water at a depth of 20–40 cm and 20–60 cm in April and May, 0–40 cm from June to August (Figure 4). For the adult shrubs, the stable-

isotope ratios ($\delta^{18}\text{O}$) of the xylem water were similar to those of the soil water at a depth of 150–300 cm

and even to the groundwater table during the measurement period (Figure 5).

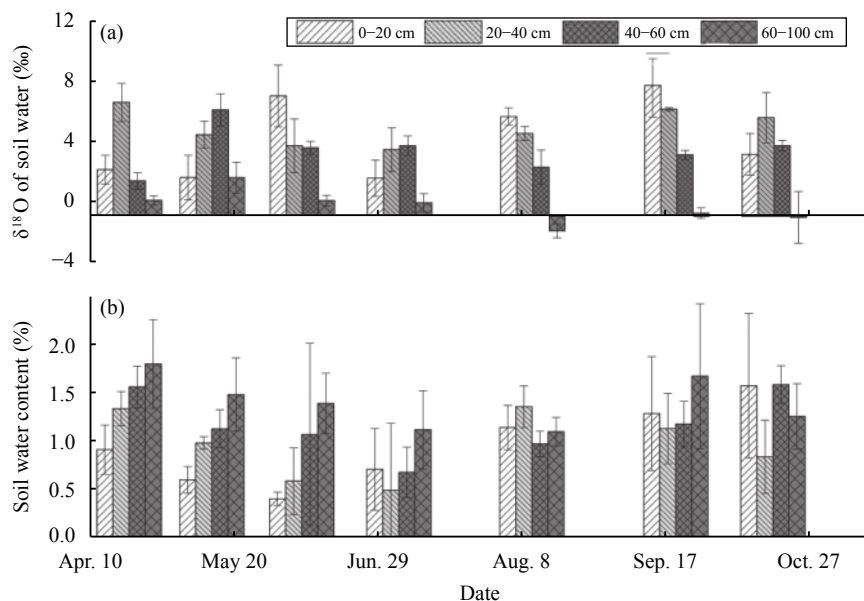


Figure 3 Seasonal variations in the stable-isotope composition of soil water (a) and soil water content (b) at depths of 0–100 cm in the study site

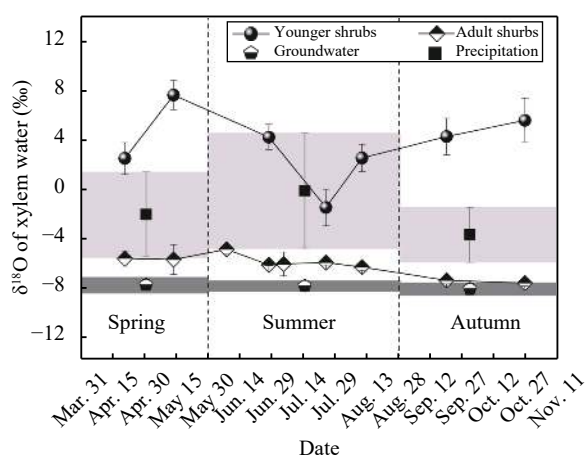


Figure 4 Stable-isotope ratios of precipitation, groundwater, and stem water of *Caragana microphylla* in the 2014 growing season. The light gray bars depict the average $\delta^{18}\text{O}$ values for precipitation; the dark gray bars depict the average $\delta^{18}\text{O}$ values for groundwater

3.4 Plant-water sources

During the measurement period, the main water source of the study species showed great season variation (Figure 5). Using the IsoSource mixing models indicated that shrubs were able to uptake water simultaneously from four potential water sources (shallow-, mid-, deep-soil water, and groundwater) but in vary-

ing relative amounts, depending on the seasonal variation of the soil moisture. For the young shrubs in spring (April and May), the contribution of shallow-soil water was in the range of 41.2%–68.3%; and the possible range of contribution by mid- and deep-soil water was 38.5%–55.9% and 0.0%–14.2%, respectively. However, the shallow-soil water contribution increased to 54.2%–78.9% in summer (June to August) as the precipitation increased, decreasing in autumn. Water sources for adult shrubs also fluctuated with the seasons. Shallow- and mid-soil water contributions were 0.0%–13.5% and 0.0%–23.8%, respectively; the deep-soil water and groundwater contributions were 32.0%–45.7% and 41.6%–69.8% in spring, respectively. During the summer, the contribution of shallow-soil water increased, while those of deep-soil water and groundwater decreased, slightly. In fact, the adult shrubs' main water sources were the deep-soil water and groundwater. So, the shrubs' water sources changed from shallow soil charged by precipitation to the more stable and abundant deep water, such as deep-soil water and groundwater as the shrub aged.

4 Discussion

4.1 Variation in stable-isotope of precipitation, soil water, and groundwater

Precipitation is a primary input for soil water and

groundwater, and impacts a plant's acquisition of resources and its survivorship in arid regions (Weltzin *et al.*, 2003; Schwinning *et al.*, 2005; Tan *et al.*, 2017). The stable-isotope ratio in precipitation is an integrated tracer of atmospheric processes worldwide and been used widely in understanding ecohydrological processes (Gat, 1996; Farquhar and Gan, 2003; Zhang and Wang, 2016). At the study site, the precipitation stable-isotope composition significantly varied

with the seasons, perhaps caused by atmospheric flow paths of vapor trajectories, different water vapor origins, or temperature and the precipitation amounts (Johnson and Ingram, 2004; Yamanaka *et al.*, 2007; Aggarwal *et al.*, 2016). The mean stable-isotope ratio of precipitation was enriched in the hot summer and depleted in spring and autumn, showing a significant temperature effect in arid regions (Guo *et al.*, 2015; Akers *et al.*, 2017).

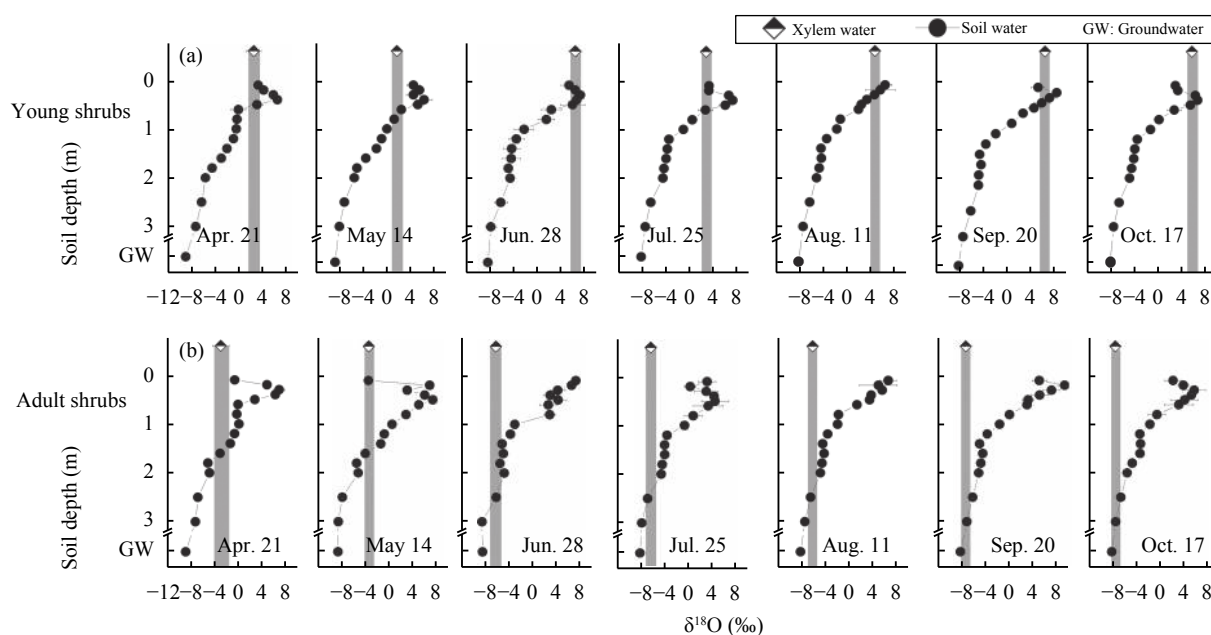


Figure 5 Stable-isotope ratios ($\delta^{18}\text{O}$) of soil water and xylem water of *Caragana microphylla* during the measurement period in 2014. Error bars represent standard errors of the mean for stable-isotope ratios ($\delta^{18}\text{O}$) for soil water, $n = 5$. The gray bars depict the average stable-isotope ratios ($\delta^{18}\text{O}$, \pm SE) for plant xylem water for each sampling date

Under the influence of seasonal precipitation inputs and the evaporation effect, the stable-isotope ratios of upper-soil water shows significant seasonal changes; however, the range of variation of isotope values is obviously less than that of precipitation because soil water is generally a mixture generated by precipitation and the moisture already in the soil (Tang and Feng, 2001; Orlowski *et al.*, 2016). Hydrologic processes—such as precipitation, evaporation, infiltration, and plant uptake and transpiration—regulated the soil-water condition (Piao *et al.*, 2009), causing clear isotopic gradients in the soil profiles (Figure 4). In early spring, low temperature and some water from ice and snow melt recharged the soil water, bringing about low stable-isotope values (Wu *et al.*, 2014; Zhou *et al.*, 2015). As time went on, the isotope values of upper-soil water were changed by rainfall and strong evaporation-caused isotopic enrichment; and the values were over about 5‰ in summer and early autumn (Figure 3a). Due to water molecules that contain light oxygen evaporating slightly more effi-

ciently than water molecules containing heavy oxygen, evaporation produces residual water enriched in the heavier isotopes, relative to the initial isotopic composition (Lee *et al.*, 2007; Song *et al.*, 2011). Because of the limited amount of summer rain that can infiltrate into deep soil, the groundwater capillary uplift is more likely to be recharged the deep soil; the SWC increases and the stable isotope of soil water decreases with depth (Figure 5). It was proposed by Zimmermann *et al.* (1967) that the effect of evaporation on stable-isotope composition of soil water is affected, with the heavy isotope enriched close to the upper layer and decreased exponentially with depth. In fact, soil water is a mixture from multiple precipitation events' infiltration, even the groundwater capillary recharge, which is the link between rainfall and groundwater (Tan *et al.*, 2017; Yang and Fu, 2017). The stable isotope composition of groundwater showed mild changes with the seasons and was isotopically depleted compared with the summer precipitation. So, the local precipitation infiltration may not

be the main source for groundwater. However, the similarity in isotopic compositions of groundwater at our site and river water indicated that the mountain river was a major contributor to recharge in the southern basin. The calculated recharge altitudes indicated that recharge water came from the Qilian Mountains (Zhou *et al.*, 2017).

4.2 Variation in plant-water sources of *C. microphylla*

Our results showed that the shrub species *C. microphylla* exhibited strong differences in sources of water uptake between the young and adult shrubs (Figures 4, 5 and 6). This pattern was consistent with the findings of several other studies, which reported that plants could shift water sources from shallow-soil water to deep water as their age increased (Song *et al.*, 2016). The stable-isotope ratio ($\delta^{18}\text{O}$) of the shrub xylem water and the IsoSource Model results showed that the young shrubs' main water sources was the upper soil water, but the adult shrubs' water utilized both soil water and groundwater, indicated that adaptive

changes in water-use patterns exists in xerophilous plants. In fact, the trees are attributed to the different root distribution, defining the depth to or volume from which plants can potentially extract water (Ehleringer *et al.*, 1991; Dawson and Pate, 1996). For the young shrubs, precipitation-derived upper-soil water is the main water source, with the limited root distribution determining the range of its water uptake. However, given the scant rain in the extremely arid region, precipitation can't meet the water requirement as the shrub's size increases. Thus the species develops not only the shallow, resource-acquiring lateral roots but also, with age, the deep-penetrating tap (sinker) roots, a common trait in desert ecosystems (Zhou *et al.*, 2017). The adult shrubs uptake mostly deep water (including deep-soil water and groundwater), providing a relatively stable water source to avoid drought stress in the desert environment. Indeed, some desert shrubs' survival in desert ecosystems has been showed to totally depend on their ability to absorb deep water by tap roots (Pate *et al.*, 1995; Canadell *et al.*, 1996; Bordron *et al.*, 2018).

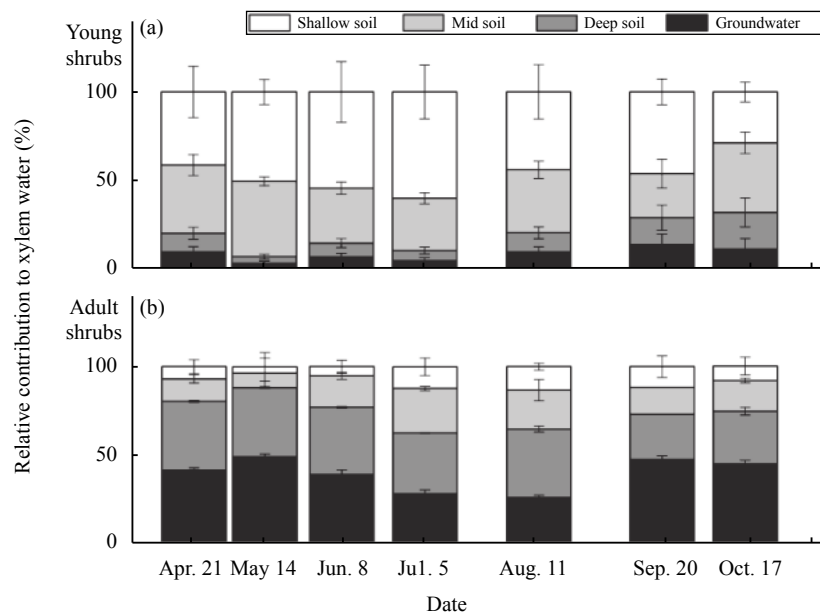


Figure 6 Seasonal changes in the relative mean contribution of soil water at different depths to *Caragana microphylla*. Data obtained from the IsoSource mixing model: shallow soil (0–50 cm), mid soil (50–150 cm), deep soil (150–300 cm), and groundwater (below 300 cm). Bars represent possible ranges of potential water sources

Isotopic values ($\delta^{18}\text{O}$) of shrub xylem water revealed that the young and adult *C. microphylla* shrub uptake of the water sources showed significantly seasonal variation (Figures 4, 5 and 6). When soil water was relatively plentiful in summer, the shrubs increased the proportion of upper-soil water absorbed, especially for young shrubs. However, when the upper-soil water became less available in spring and autumn, the shrubs increased the proportion of deeper-

soil water absorbed, especially for adult shrubs, for which the main water sources were the deep-soil water and groundwater. Many studies have demonstrated seasonal variations in utilization of water sources, with most of their transpiration water derived from shallow layers under wet conditions and more from deep-soil layers or groundwater under dry conditions (Dawson and Pate, 1996; Wu *et al.*, 2014; Song *et al.*, 2016). Especially in dry conditions, the

shift of the main water sources from shallow layers to deep layers appears to be a very important survival strategy for shrubs in arid desert lands (Dawson and Pate, 1996).

C. microphylla, generally planted in sand dunes for decreasing wind speed and intercepting drift sand, had limited use of shallow-soil water once it grew into an adult shrub (Figure 6). Although plants could use reliable summer precipitation, as the upper-soil water condition had been improved by the precipitation input (Ehleringer and Dawson, 1992), the adult shrubs' main water sources were the deep-soil water and groundwater. A combination of deep-soil water and groundwater has been recognized as a stable and reliable water source, although more than 80% of the rainfall occurred during summer in the study area; the shallow-soil water maintains only more shallow and short-lived soil-moisture resources due to high evaporative demand (Dai *et al.*, 2015; Zhou *et al.*, 2017). The results revealed that upper-soil water was an unreliable water source, especially for some adult shrubs in an arid region. So, with age, shrubs exploit deeper water sources for long-term survival and sustainable development.

5 Conclusions

In this study, we used the stable-isotope ratio ($\delta^{18}\text{O}$) for detecting water sources of young and adult *C. microphylla* species. The results indicated that xylem water $\delta^{18}\text{O}$ values exhibited a significant difference between the young and adult shrubs, suggesting that the shrub absorbed different water sources at different ages. According to the results of the IsoSource model, the young shrubs took up 41.2%–68.3% of their water from shallow-soil water, with the variation determined by the seasonality of precipitation inputs; but the adult shrubs took up 41.6%–69.8% of their water from groundwater, with very limited seasonal variation. Namely, the water-use patterns of *C. microphylla* exhibited significant variation with the age. For the young shrubs, upper-soil water recharged by precipitation was the main water source; and it has a significant seasonal variation with the seasonal variation in precipitation inputs. However, the main water sources for adult shrubs were the deep-soil layers and groundwater. Therefore, from a long-term perspective, this water-use strategy of *C. microphylla* could be an advantage in the competition for the scarce water resources in desert ecosystem.

Acknowledgments:

This study was supported by the National Science Foundation for Distinguished Young Scholars of China (Grant No. 41701035), the Key Program of National Natural Science Foundation of China (Grant

No. 41630861), and the National Science Foundation for Post-doctoral Scientists of China (Grant No. 2016M602902). The authors are very grateful to the anonymous reviewers and editors for their critical reviews and comments that helped to improve and clarify the manuscript.

References:

- Aggarwal PK, Romatschke U, Araguasaraguas L, *et al.*, 2016. Proportions of convective and stratiform precipitation revealed in water isotope ratios. *Nature Geoscience*, 9(8): 624–629. DOI: [10.1038/NGEO2739](https://doi.org/10.1038/NGEO2739).
- Akers PD, Welker JM, Brook GA, 2017. Reassessing the role of temperature in precipitation oxygen isotopes across the eastern and central united states through weekly precipitation-day data. *Water Resources Research*, 53(9): 7644–7661. DOI: [10.1002/2017WR020569](https://doi.org/10.1002/2017WR020569).
- Barbeta A, Mejia-Chang M, Ogaya R, *et al.*, 2015. The combined effects of a long-term experimental drought and an extreme drought on the use of plant-water sources in a Mediterranean forest. *Global Change Biology*, 21(3): 1213–1225. DOI: [10.1111/gcb.12785](https://doi.org/10.1111/gcb.12785).
- Bordon B, Robin A, Oliveira IR, *et al.*, 2018. Fertilization increases the functional specialization of fine roots in deep soil layers for young *Eucalyptus grandis* trees. *Forest Ecology and Management*, 431: 6–16. DOI: [10.1016/j.foreco.2018.03.018](https://doi.org/10.1016/j.foreco.2018.03.018).
- Bunker DE, DeClerck F, Bradford JC, *et al.*, 2005. Species loss and aboveground carbon storage in a tropical forest. *Science*, 310(5750): 1029–1031. DOI: [10.1126/science.1117682](https://doi.org/10.1126/science.1117682).
- Canadell J, Jackson RB, Ehleringer JR, *et al.*, 1996. Maximum rooting depth of vegetation types at the global scale. *Oecologia*, 108: 583–595. DOI: [10.1007/BF00329030](https://doi.org/10.1007/BF00329030).
- Dai Y, Zheng XJ, Tang LS, *et al.*, 2015. Stable oxygen isotopes reveal distinct water use patterns of two *Haloxylon* species in the Gurbantonggut Desert. *Plant and Soil*, 389(1–2): 73–87. DOI: [10.1007/s11104-014-2342-z](https://doi.org/10.1007/s11104-014-2342-z).
- Dansgaard W, 1964. Stable isotopes in precipitation. *Tellus*, 16: 436–468. DOI: [10.3402/tellusa.v16i4.8993](https://doi.org/10.3402/tellusa.v16i4.8993).
- Dawson TE, Pate JS, 1996. Seasonal water uptake and movement in root systems of Australian phraeatophytic plants of dimorphic root morphology: a stable isotope investigation. *Oecologia*, 107(1): 13–20. DOI: [10.1007/BF00582230](https://doi.org/10.1007/BF00582230).
- Dawson TE, Mambelli S, Plamboeck AH, *et al.*, 2002. Stable isotopes in plant ecology. *Annual Review of Ecology and Systematics*, 33: 507–559. DOI: [10.1146/annurev.ecolsys.33.020602.095451](https://doi.org/10.1146/annurev.ecolsys.33.020602.095451).
- Donovan LA, Ehleringer JR, 1991. Ecophysiological differences among juvenile and reproductive plants of several woody species. *Oecologia*, 86(4): 594–597. DOI: [10.1007/BF00318327](https://doi.org/10.1007/BF00318327).
- Drake PL, Froend RH, Franks PJ, 2011. Linking hydraulic conductivity and photosynthesis to water-source partitioning in trees versus seedlings. *Tree Physiology*, 31(7): 763–773. DOI: [10.1093/treephys/tpr068](https://doi.org/10.1093/treephys/tpr068).
- Eggemeyer KD, Awada T, Harvey FE, *et al.*, 2009. Seasonal changes in depth of water uptake for encroaching trees *Juniperus virginiana* and *Pinus ponderosa* and two dominant C_4 grasses in a semiarid grassland. *Tree Physiology*, 29(2): 157–169. DOI: [10.1093/treephys/tpn019](https://doi.org/10.1093/treephys/tpn019).
- Ehleringer JR, Dawson TE, 1992. Water uptake by plants: perspectives from stable isotope composition. *Plant, Cell & Environment*, 15(9): 1073–1082. DOI: [10.1111/j.1365-3040.1992.tb01657.x](https://doi.org/10.1111/j.1365-3040.1992.tb01657.x).

- Ehleringer JR, Phillips SL, Schuster WSF, *et al.*, 1991. Differential utilization of summer rains by desert plants. *Oecologia*, 88(3): 430–434. DOI: [10.1007/BF00317589](https://doi.org/10.1007/BF00317589).
- Ehleringer JR, Roden J, Dawson TE, 2000. Assessing ecosystem level water relations through stable isotope analyses. In: O. E. Sala, R. B. Jackson, H. A. Mooney, & R. W. Howarth (Eds.), (eds) *Methods in ecosystem science*. (pp. 181–198). Berlin, Heidelberg, New York: Springer.
- Ellsworth PZ, Williams DG, 2007. Hydrogen isotope fractionation during water uptake by woody xerophytes. *Plant and Soil*, 291(1–2): 93–107. DOI: [10.1007/s11104-006-9177-1](https://doi.org/10.1007/s11104-006-9177-1).
- Fay PA, Carlisle JD, Knapp AK, *et al.*, 2003. Productivity responses to altered rainfall patterns in a C₄-dominated grassland. *Oecologia*, 137(2): 245–251. DOI: [10.1007/s00442-003-1331-3](https://doi.org/10.1007/s00442-003-1331-3).
- Farquhar GD, Gan KS, 2003. On the progressive enrichment of the oxygen isotopic composition of water along a leaf. *Plant, Cell & Environment*, 26(6): 801–819. DOI: [10.1046/j.1365-3040.2003.01013.x](https://doi.org/10.1046/j.1365-3040.2003.01013.x).
- Garcia-Forner N, Adams HD, Sevanto S, 2016. Responses of two semiarid conifer tree species to reduced precipitation and warming reveal new perspectives for stomatal regulation. *Plant, Cell & Environment*, 39(1): 38–49. DOI: [10.1111/pce.12588](https://doi.org/10.1111/pce.12588).
- Gay JR, 1996. Oxygen and hydrogen isotopes in the hydrologic cycle. *Annual Review of Earth and Planetary Sciences*, 24: 225–262. DOI: [10.1146/annurev.earth.24.1.225](https://doi.org/10.1146/annurev.earth.24.1.225).
- Grossiord C, Sevanto S, Dawson TE, *et al.*, 2017. Warming combined with more extreme precipitation regimes modifies the water sources used by trees. *New Phytologist*, 213(2): 584–596. DOI: [10.1111/nph.14192](https://doi.org/10.1111/nph.14192).
- Guan PT, Zhang XK, Yu J, *et al.*, 2015. Variation of soil nematode community composition with increasing sand-fixation year of *Caragana microphylla*: Bioindication for desertification restoration. *Ecological Engineering*, 81: 93–101. DOI: [10.1016/j.ecoleng.2015.04.011](https://doi.org/10.1016/j.ecoleng.2015.04.011).
- Guo XY, Feng Q, Wei YP, *et al.*, 2015. An overview of precipitation isotopes over the Extensive Hexi Region in NW China. *Arabian Journal of Geosciences*, 8(7): 4365–4378. DOI: [10.1007/s12517-014-1521-9](https://doi.org/10.1007/s12517-014-1521-9).
- Johnson KR, Ingram BL, 2004. Spatial and temporal variability in the stable isotope systematics of modern precipitation in china: implications for paleoclimate reconstructions. *Earth and Planetary Science Letters*, 220(3–4): 365–377. DOI: [10.1016/S0012-821X\(04\)00036-6](https://doi.org/10.1016/S0012-821X(04)00036-6).
- Kendall C, McDonnell JJ, 1998. *Isotope tracers in catchment hydrology*. New York: Elsevier Sci.
- Kerhoulas LP, Kolb TE, Koch GW, 2013. Tree size, stand density, and the source of water used across seasons by ponderosa pine in northern Arizona. *Forest Ecology and Management*, 289: 425–433. DOI: [10.1016/j.foreco.2012.10.036](https://doi.org/10.1016/j.foreco.2012.10.036).
- Lee KS, Kim JM, Lee DR, *et al.*, 2007. Analysis of water movement through an unsaturated soil zone in Jeju Island, Korea using stable oxygen and hydrogen isotopes. *Journal of Hydrology*, 345: 199–211. DOI: [10.1016/j.jhydrol.2007.08.006](https://doi.org/10.1016/j.jhydrol.2007.08.006).
- Lilley JM, Fukai S, 1994. Effect of timing and severity of water deficit on four diverse rice cultivars I. Rooting pattern and soil water extraction. *Field Crops Res*, 37: 205–213. DOI: [10.1016/0378-4290\(94\)90099-X](https://doi.org/10.1016/0378-4290(94)90099-X).
- Loik ME, Breshears DD, Lauenroth WK, *et al.*, 2004. A multi-scale perspective of water pulses in dry land ecosystems: climatology and ecohydrology of the western USA. *Oecologia*, 141: 269–281. DOI: [10.1007/s00442-004-1570-y](https://doi.org/10.1007/s00442-004-1570-y).
- Matzner SL, Rice KJ, Richards JH, 2003. Patterns of stomatal conductance among blue oak (*Quercus douglasii*) size classes and populations: implications for seedling establishment. *Tree Physiology*, 23(11): 777–784. DOI: [10.1093/treephys/23.11.777](https://doi.org/10.1093/treephys/23.11.777).
- Mensforth LJ, Thorburn PJ, Tyerman SD, *et al.*, 1994. Sources of water used by riparian Eucalyptus camaldulensis overlying highly saline groundwater. *Oecologia*, 100: 21–28. DOI: [10.1007/BF00317126](https://doi.org/10.1007/BF00317126).
- Orlowski N, Pratt DL, McDonnell JJ, 2016. Intercomparison of soil pore water extraction methods for stable isotope analysis. *Hydrological Processes*, 30(19): 3434–3449. DOI: [10.1002/hyp.10870](https://doi.org/10.1002/hyp.10870).
- Pate JS, Jeschke WD, Aylward MJ, 1995. Hydraulic architecture and xylem structure of the dimorphic root systems of South-West Australian species of Proteaceae. *Journal of Experimental Botany*, 46(8): 907–915. DOI: [10.1093/jxb/46.8.907](https://doi.org/10.1093/jxb/46.8.907).
- Phillips DL, Gregg JW, 2003. Source partitioning using stable isotopes: Coping with too many sources. *Oecologia*, 136: 261–269. DOI: [10.1007/s00442-003-1218-3](https://doi.org/10.1007/s00442-003-1218-3).
- Piao SL, Yin L, Wang XH, *et al.*, 2009. Summer soil moisture regulated by precipitation frequency in China. *Environmental Research Letters*, 4(4): 044012. DOI: [10.1088/1748-9326/4/4/044012](https://doi.org/10.1088/1748-9326/4/4/044012).
- Rossatto DR, Silva LDCR, Villalobos-Vega R, *et al.*, 2012. Depth of water uptake in woody plants relates to groundwater level and vegetation structure along a topographic gradient in a neotropical savanna. *Environmental and Experimental Botany*, 77: 259–266. DOI: [10.1016/j.envexpbot.2011.11.025](https://doi.org/10.1016/j.envexpbot.2011.11.025).
- Schultz NM, Griffis TJ, Lee XH, *et al.*, 2011. Identification and correction of spectral contamination in ²H/¹H and ¹⁸O/¹⁶O measured in leaf, stem, and soil water. *Rapid Communications in Mass Spectrometry*, 25(21): 3360–3368. DOI: [10.1002/rcm.5236](https://doi.org/10.1002/rcm.5236).
- Schwinning S, Ehleringer JR, 2001. Water use trade offs and optimal adaptations to pulse-driven arid ecosystems. *Journal of Ecology*, 89: 464–480. DOI: [10.1046/j.1365-2745.2001.00576.x](https://doi.org/10.1046/j.1365-2745.2001.00576.x).
- Schwinning S, Starr BI, Ehleringer JR, 2005. Summer and winter drought in a cold desert ecosystem (Colorado Plateau) part I: Effects on soil water and plant water uptake. *Journal of Arid Environments*, 60(4): 547–566. DOI: [10.1016/j.jaridenv.2004.07.003](https://doi.org/10.1016/j.jaridenv.2004.07.003).
- Snyder KA, Williams DG, 2003. Defoliation alters water uptake by deep and shallow roots of *Prosopis velutina* (Velvet Mesquite). *Functional Ecology*, 17(3): 363–374. DOI: [10.1046/j.1365-2435.2003.00739.x](https://doi.org/10.1046/j.1365-2435.2003.00739.x).
- Song XF, Wang P, Yu JJ, *et al.*, 2011. Relationships between precipitation, soil water and groundwater at Chongling catchment with the typical vegetation cover in the Taihang mountainous region, China. *Environment and Earth Science*, 62: 787–796. DOI: [10.1007/s12665-010-0566-7](https://doi.org/10.1007/s12665-010-0566-7).
- Song LN, Zhu JJ, Li MC, *et al.*, 2016. Water use patterns of *Pinus sylvestris* var. *mongolica* trees of different ages in a semiarid sandy lands of Northeast China. *Environmental and Experimental Botany*, 129: 94–107. DOI: [10.1016/j.envexpbot.2016.02.006](https://doi.org/10.1016/j.envexpbot.2016.02.006).
- Su YZ, Zhao HL, 2003. Soil properties and plant species in an age sequence of *Caragana microphylla* plantations in the Horqin Sandy Land, north China. *Ecological Engineering*, 20(3): 223–235. DOI: [10.1016/S0925-8574\(03\)00042-9](https://doi.org/10.1016/S0925-8574(03)00042-9).
- Tan HB, Liu ZH, Rao WB, *et al.*, 2017. Stable isotopes of soil water: Implications for soil water and shallow groundwater recharge in hill and gully regions of the Loess Plateau, China. *Agriculture, Ecosystems & Environment*, 243: 1–9. DOI: [10.1016/j.agee.2017.04.001](https://doi.org/10.1016/j.agee.2017.04.001).
- Tang KL, Feng XH, 2001. The effect of soil hydrology on the oxygen and hydrogen isotopic compositions of plants' source water. *Earth*

- and Planetary Science Letters, 185: 355–367. DOI: [10.1016/S0012-821X\(00\)00385-X](https://doi.org/10.1016/S0012-821X(00)00385-X).
- Trogisch S, Salmon Y, He JS, *et al.*, 2016. Spatio-temporal water uptake patterns of tree saplings are not altered by interspecific interaction in the early stage of a subtropical forest. *Forest Ecology and Management*, 367: 52–61. DOI: [10.1016/j.foreco.2016.02.018](https://doi.org/10.1016/j.foreco.2016.02.018).
- Weltzin JF, Loik ME, Schwinning S, *et al.*, 2003. Assessing the response of terrestrial ecosystems to potential changes in precipitation. *Bioscience*, 53(10): 941–952. DOI: [10.1641/0006-3568\(2003\)053\[0941:ATROTE\]2.0.CO;2](https://doi.org/10.1641/0006-3568(2003)053[0941:ATROTE]2.0.CO;2).
- West AG, Sperry JS, Bush SE, *et al.*, 2008. Transpiration and hydraulic strategies in a pinon-juniper woodland. *Ecol Appl*, 18: 911–927. DOI: [10.1890/06-2094.1](https://doi.org/10.1890/06-2094.1).
- Wu Y, Zhou H, Zheng XJ, *et al.*, 2014. Seasonal changes in the water use strategies of three co-occurring desert shrubs. *Hydrological Processes*, 28(26): 6265–6275. DOI: [10.1002/hyp.10114](https://doi.org/10.1002/hyp.10114).
- Xu H, Li Y, 2006. Water-use strategy of three Central Asian desert shrubs and their responses to rain pulse events. *Plant and Soil*, 285: 5–17. DOI: [10.1007/s11104-005-5108-9](https://doi.org/10.1007/s11104-005-5108-9).
- Yamanaka T, Tsujimura M, Oyunbaatar D, *et al.*, 2007. Isotopic variation of precipitation over eastern Mongolia and its implication for the atmospheric water cycle. *Journal of Hydrology*, 333: 21–34. DOI: [10.1016/j.jhydrol.2006.07.022](https://doi.org/10.1016/j.jhydrol.2006.07.022).
- Yang YG, Fu BJ, 2017. Soil water migration in the unsaturated zone of semiarid region in china from isotope evidence. *Hydrology and Earth System Sciences*, 21(3): 1757–1767. DOI: [10.5194/hess-21-1757-2017](https://doi.org/10.5194/hess-21-1757-2017).
- Zhang MJ, Wang SJ, 2016. A review of precipitation isotope studies in China: basic pattern and hydrological process. *Journal of Geographical Sciences*, 26(7): 921–938. DOI: [10.1007/s11442-016-1307-y](https://doi.org/10.1007/s11442-016-1307-y).
- Zhou H, Zhao WZ, Zheng XJ, *et al.*, 2015. Root distribution of *Nitraria sibirica* with seasonally varying water sources in a desert habitat. *Journal of Plant Research*, 128(4): 613–622. DOI: [10.1007/s10265-015-0728-5](https://doi.org/10.1007/s10265-015-0728-5).
- Zhou H, Zhao WZ, Zhang GF, 2017. Varying water utilization of *Haloxylon ammodendron* plantations in a desert-oasis ecotone. *Hydrological Processes*, 31(4): 825–835. DOI: [10.1002/hyp.11060](https://doi.org/10.1002/hyp.11060).
- Zimmermann U, Munnich KO, Roether W, 1967. In: Glenn, E.S. (Ed.), *Downward Movement of Soil Moisture Traced by Means of Hydrogen Isotopes*, *Isotope Techniques in the Hydrologic Cycle*, vol. 11. American Geophysical Union, the Geophysical Monograph, pp. 28–36.